

Collaborative exploration of genetic resources of papaya and mountain papayas for their improvement and promotion

Colombia: Geo Coppens d'Eeckenbrugge¹, John A. Ocampo¹, Creuci M. Caetano¹, Daniel Jimenez⁴, Cristian A. Olaya², Maria T. Restrepo⁴, Carlos Reyes⁵, Rodrigo Hoyos⁵, Mario Lobo³, Clara I. Medina³, Francisco Morales², Alba M. Torres²; Costa Rica: Eric Mora⁶; Ecuador: Jorge Vega⁷, Lenin Paz⁸; Francia: Patrick Ollitrault¹², Fabrice Le Bellec¹²; Venezuela: Carolina Rosales⁹, Ariadne Vegas⁹, Dilia Rodriguez¹⁰, Anna Maselli⁹, Edgoris Marys¹¹

¹CIRAD-FLHOR/IPGRI, ²CIAT, ³CORPOICA, ⁴Univ. de Caldas, ⁵Univ. Nacional de Colombia, ⁶Univ. de Costa Rica, ⁷Univ. Técnica de Ambato, ⁸INIAP, ⁹INIA/CENIAP Venezuela, ¹⁰MARN/CNRP, ¹¹IVIC/MCT, ¹²CIRAD

Objectives

The objective was to strengthen and integrate regional efforts to find solutions to the major limiting factors of papaya cultivation in the Andean and Caribbean countries, collecting, conserving and studying the variability of the cultivated *Caricaceae* and their wild relatives as well as the variability of their main pathogens in the participating countries, in an effort to develop genetic resistance through the application of traditional and biotechnological methods.

Activities

- Germplasm collecting and conservation.
- Germplasm characterization and diversity studies.
- Selection of elite native genotypes elite for breeding.
- Sanitation and dissemination of elite native genotypes for direct use (babaco).
- Introduction of resistance genes in native or allochthonous commercial cultivars through a first cycle of intra- and interspecific crosses.
- Study of the variability of the principal pathogens and pests in the participating countries: papaya ringspot virus (PRSV), bacterial canker, anthracnose, and nematodes.
- Development of marker assisted introgression to introduce resistance/tolerance to the bacteriosis (*Erwinia* sp.) and/or PRSV.
- Training of regional scientists in taxonomy, germplasm management, bacteriology, and biotechnology



Methodology

- Training in papaya taxonomy and germplasm management.
- Collecting, recovery, exchange, and conservation of papaya genetic resources, including a basic study on seed storage.
- Botanical and agromorphological characterization and evaluation of germplasm field collections.
- Evaluation of susceptibility to bacterial canker (*Erwinia* sp.), virus and nematodes, including study of plant-host interaction.
- Isozyme and molecular characterization (PCR-RFLP, SSR).
- Cytogenetic and palynological characterization.
- Interspecific hybridizations and backcrosses.
- Study of the variability of the two main pathogens (PRSV, *Erwinia* sp.).

Overall results

- Four national field collections, with a total of 377 accessions, characterized morphologically *in situ* and/or *ex situ*. *In situ* morphological characterization of 46 accessions in Costa Rica.
- Characterization of 147, 69, and 68 accessions by isozyme, microsatellite and PCR-RFLP markers respectively.
- Cytogenetic and palynological characterization of 21 accessions from seven species.



- Inventory of the strains of PRSV and *Erwinia* in the participating countries, and study of their variability.

- National scientists trained in taxonomy, germplasm management, bacteriology, and biotechnology.



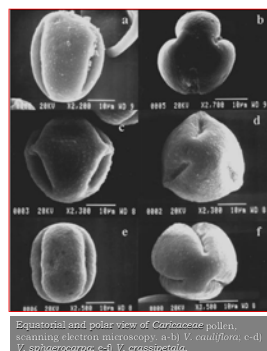
Rescue of hybrid embryos (C. papaya x V. cauliflora)



Cryopreservation of seeds



Carica papaya



Equatorial and polar view of *Caricaceae* pollen, scanning electron microscopy. a-b) *V. cauliflora*; c-d) *V. goudotiana*; e-f) *V. crassipetala*.

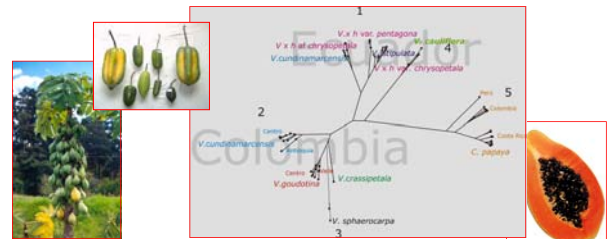
- Identification of sources of resistance against PRSV in *V. cundinamarcensis* and *V. cauliflora* and against *Erwinia* sp. in *V. cundinamarcensis* and *V. goudotiana*.
- Obtention of interspecific hybrids and identification of elite genotypes for papaya and babaco.

New knowledge and technology generated

The seed storage conditions have been defined for *ex situ* conservation of germplasm collections.

A list of morphological descriptors has been developed, allowing significant progress in our comprehension of the diversity of genetic resources of the cultivated *Caricaceae*. In particular, an unexpected heterogeneity has been observed in babaco and baby-babaco, which should be taken into account in breeding and promotion of these fruits. In Costa Rica, *in situ* morphological characterization has shown differences between wild and cultivated forms.

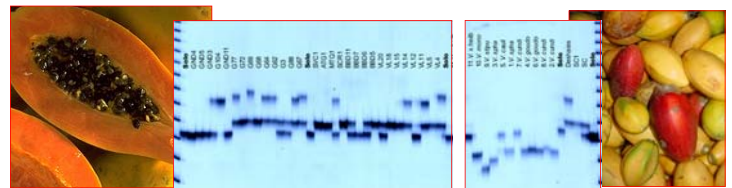
The isozyme and molecular markers have clearly differentiated the genera *Carica* and *Vasconcellea*, confirming the recent taxonomic reorganization.



Structure of isozyme diversity in the genera *Carica* and *Vasconcellea* in Colombia and Ecuador.

The isozymes have confirmed the morphological observations in *Vasconcellea*. In addition they have shown an important genetic variation in *C. papaya* in Colombia, with an important geographic component, and a strong differentiation among the accessions of *V. cundinamarcensis* of Colombia and Ecuador, with an important phenomenon of reciprocal introgression with *V. stipulata* in the second country.

The development of microsatellite markers has opened important prospects for breeding (in particular for marker assisted selection). These markers have exhibited much more polymorphism than the previously tested molecular markers (RAPD and AFLP). They have permitted a study of papaya genetic diversity in the Caribbean, which showed the existence of a geographical structure and confirmed the strong diversity of *C. papaya* in Colombia.



Microsatellite markers in the genera *Carica* and *Vasconcellea*.

The PCR-RFLP technique was developed to study the diversity of chloroplast DNA in the *Caricaceae*. In the near future, its utilization will improve our comprehension of the phylogenetic relations among species. The cytogenetic and palynological characterizations have also contributed new elements for the study of these relations.

Sources of resistance to nematodes, PRSV and *Erwinia*, potentially useful in papaya breeding, have been identified in *Vasconcellea*. These resistances can be transferred by biotechnological techniques of *in vitro* embryo culture, but the poor development of the interspecific hybrids remains an important difficulty for their introgression in the common papaya.

The joint analysis of virus samples from Venezuela, Colombia and Ecuador, as well as other parts of the world, showed that the PRSV isolates from Colombia and Ecuador are very similar to those from Mexico, Florida, and Hawaii. On the contrary, a wide genetic variability was observed among the Venezuelan isolates. These results suggest that the adoption of control techniques, as cross protection or the use of transgenic cultivars, could follow a similar strategy in Colombia and Ecuador and a few regions of Venezuela. In the other regions of this country, the potential of these control techniques looks much more limited.

Diffusion of results

The project has generated 27 publications and communications, and given opportunities for eight student theses at the graduate and postgraduate levels. Several other papers are being prepared.

Publications

- Proceedings**
Memorias del Taller Internacional sobre *Caricaceae*. Eds. científicos: F. Leal y G. Coppens d'Eeckenbrugge. FONTAGRO-IICA. Impresora Feriva, Cali, 2003.
- Journals**
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Rosales, L.C. y Suárez H., Z. 2001. Importancia de nematodos asociados al lechoso y distribución geográfica en Venezuela. Fitopatología Venezolana 14: 21-23.
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Vegas, A. G., Trujillo, J., Mata, L., Castro, M., Martínez y G. Robles. 2003. Obtención, regeneración y evaluación de híbridos intergenéricos entre *Carica papaya* y *Vasconcellea (Carica) cauliflora*. Interiencia (en revisión).
- Theses**
Benítez, S.P. 2003. Estudios de germinación de la semilla de papayuela *Vasconcellea cundinamarcensis (pubescens)* y *Vasconcellea goudotiana*. Tesis para optar al título de Ingeniero Agrónomo. Universidad Nacional Medellín.
Cadavid, A.C., Villegas, E. 2001. Evaluación y caracterización morfológicas de *Caricaceae* de altura. Tesis para optar al título de Ingenierías Agrónomas, Universidad Nacional, Medellín, Colombia.
Cadavid A.C. 2003. Caracterización isoenzimática de *Caricaceae* de altura. Tesis para optar al título de M.Sc., Universidad Nacional, Medellín, Colombia. En ejecución.
Cardozo, J. 2002. Evaluación de materiales resistentes a la marchitez bacteriana en Lechosa (*Carica papaya* L.). Tesis para optar al título de Ingeniero Agrónomo. Universidad Central de Venezuela.
Jiménez, D. 2002. Caracterización y estudio de la diversidad genética en los géneros *Vasconcellea* y *Carica* (*Caricaceae*) en Colombia y Ecuador por medio de marcadores isoenzimáticos. Tesis para optar al título de Ingeniero Agrónomo. Universidad de Caldas.
Ocampo, J.A. 2002. Développement de marqueurs microsatellites pour l'analyse du génome dans les genres *Carica* et *Vasconcellea (Caricaceae)*. Rapport pour l'obtention du Certificat d'Etudes Supérieures en Agronomie. ENSAM, Université Montpellier II.
Ocampo, J.A. 2003. Marqueurs microsatellites chez *Carica papaya* L.: développement, transférabilité et étude de la diversité génétique Mémoire présenté pour l'obtention du Diplôme d'Etudes Approfondies (equ. Master), spécialité Ressources Génétiques et Interactions Biologiques. ENSAM, Université Montpellier II.
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